## CHARACTERIZATION OF SCLEROTINIA SCLEROTIORUM ISOLATES USED IN COMMON BEAN SCREENING FROM BEAN PRODUCTION AREAS IN THE UNITED STATES

L. K. Otto-Hanson, K. Powers, J. R. Steadman, Department of Plant Pathology, University of Nebraska-Lincoln

There is a dearth of information on variability of isolates of *Sclerotinia sclerotiorum* from common bean. In addition, variability between data from collaborators screening for putative resistance has been reported (Steadman et al, this issue). Pathogen isolates were collected from collaborators from locations within the U.S. bean production areas to determine mycelial compatibility groupings (MCGs), aggressiveness, and molecular genotype.

Sclerotia of nine *S. sclerotiorum* isolates used for screening in greenhouses/labs were collected from the following U.S. sites: ID, ND, WI, WA, NE, CO, MI, OR, and NY. These nine isolates were tested for clonality using MCGs (Kull et al, 2004). One 8mm plug of one isolate was placed on one side of a plate of red PDA (Potato Dextrose Agar) and another plug of the same or a different isolate was placed 2.5 cm across from it. After 7-14 days, if the isolates formed a barrage line where the hyphae from the isolates met, then the isolates were incompatible and were considered unique isolates. If there was no evidence of mycelial interaction, the isolates were compatible, and were considered clones. The nine isolates were tested in a matrix for MCGs (Table 1). No two greenhouse/lab isolates were compatible; each isolate was unique.

The nine isolates were also tested for aggressiveness using the detached leaf test (DLT) (Steadman et al, 1997). In the DLT, a detached trifoliolate leaf was inoculated with an 8 mm agar plug containing *S. sclerotiorum* mycelium. After 48 hours of incubation in high humidity, the area of the lesion caused by the pathogen was measured by digital analysis. The most aggressive isolates used in greenhouse/lab screening were from ID, ND, and WI; these isolates were significantly more aggressive than greenhouse/lab screening isolates from NE, WA, and CO (Table 2).

We have shown that the internal transcribed spacer region (nuclear subunit rDNA) can be used for interspecific separation of *S. sclerotiorum*, *S. trifoliorum*, and *S. minor*. A preliminary study of the nuclear large subunit rDNA failed to find intraspecific variation for the nine screening isolates. Further investigation of this region as well as molecular characterization by the use of L. Kohn's microsatellites is planned to determine if polymorphisms found by molecular genotyping are related to aggressiveness of the isolates.

The variation in aggressiveness and MCGs may help explain why greenhouse/lab screening results often do not agree across different test sites. When collaborators described greenhouse/lab screening protocols, such as the straw test, variations in each protocol such as stage of plant inoculation, apex vs. petiole could also have contributed to variation in results across test sites. The selection of a universal isolate(s) for use in all screening tests may result in more consistent similarity in ranking of white mold resistance sources.

Table. 1. Results of the MCG test of the nine greenhouse Sclerotinia sclerotiorum bean isolates.

	NY	СО	WA	WI	ND	MI	OR	NE	ID
NY	0	X	X	X	X	X	X	X	X
СО		О	X	X	X	X	X	X	X
WA			0	X	X	X	X	X	X
WI				0	X	X	X	X	X
ND					О	X	X	X	X
MI						0	X	X	X
OR					-		О	X	X
NE								0	X
ID									О

<sup>(</sup>O) = Compatible MCG reaction; (X) = Incompatible MCG reaction

Table 2. Mean lesion size of nine *Sclerotinia sclerotiorum* isolates on bean line G122 using the detached leaf test.

ISOLATE-SOURCE	MEAN LESION SIZE	t GROUPING			
JRS 483-IDAHO	17.5	A			
JRS 274-NORTH DAKOTA	17.3	A			
JRS 478-WISCONSIN	17.0	A			
JRS 443-NEW YORK	15.8	A	В		
JRS 482-MICHIGAN	15.7	A	В	C	
JRS 455-OREGON	14.2	A	В	C	
JRS 152-NEBRASKA	12.8		В	C	
JRS 467-COLORADO	12.0			С	
JRS 456-WASHINGTON	8.3	D			

LSD=3.14; ALPHA=0.05

## References

Kull, L.S., Pedersen, W.L., Palmquist, D. and Hartman, G.L. 2004. Plant Dis. 88:325-332. Steadman, J.R., Powers, K., and Higgins, B. 1997. Ann Rep. Bean Improv. Coop. 40:140-141.